Protocol for Rearing Glassy-Winged Sharpshooter

Field Collected Rearing Techniques:

- 1. Collect insects when the weather is cool, no greater than 62-65 F (17-18 C), at higher temperatures GWSS begin to fly making collection difficult.
- 2. The best collection is in untreated commercial groves of citrus, though collections can also be made from windbreak plantings, native vegetation around citrus groves and ornamental plantings in urban areas.
- 3. Using the field collection technique, insects are captured by placing a canvas sweep net under a branch with GWSS and using a stick to knock the insects off the branches.
- 4. Insects are transported in collection cages containing a feeding plant or a branch from the field to the lab where they are separated from plant material and other insects with an aspirator.
- 5. After aspirating into vials of 100 insects, a second cleaning using Co2 removes any smaller, unwanted insects from GWSS vials.
- 6. Insects are then counted, documented and added to field collection cages.
- 7. Field collection cages should be set up with 2 cowpea plants and 2 euonymus plants.
- 8. Each cage should get 300 insects per four plants.

Lab Rearing Techniques:

- 1. After leaves with egg masses have been removed from the plant, carefully trim leaf so it will fit into petri dish leaving as much of the leaf as possible.
- 2. Place cotton pad (Johnson's Pure cotton cosmetic rounds) into petri dish and dampen with water using Nalgene wash bottle. Place egg mass on top of cotton pad and cover with lid.
- 3. Place petri dishes into plastic tray and put into incubator. 80% humidity, 27 C, 16 hour day.
- 4. Check petri dishes daily for emerged nymphs and dry cotton pads. If pad is dry add a few drops of water. Do not add too much water or nymphs will drown.

- 5. Release emerged nymphs into nymph cages by gently setting petri dishes inside cage in greenhouse containing 4 feeding plants, remove lids after all dishes have been placed. No more than 300 nymphs per cage.
- 6. Greenhouse settings for nymphs should be 16:8, L:D Photoperiod, high temperature 90, and low temperature 72.
- 7. Try to not disturb nymphs for the first month. If a plant needs to be changed carefully cut plant and lean stalk of cut plant onto pot of new plant. Remove cut plant as soon as nymphs have moved to new plant.
- 8. At 2nd instar remove one cowpea and add 1 sunflower.(min.6")
- 9. After one month, or when insects have become adults, remove chrysanthemum and add 1 cowpea.(Cage should have 2 cowpeas, 1 Sunflower, 1 Oviposition plant)
- 10. After adults start to darken in color from their initial reddish hue about (12 to 14 days after becoming adults). Check cages daily for gravid females. (Gravid females will have 2 white spots on either side of their lower abdomen. Remove sunflower and replace with oviposition plant (2 cowpeas, 2 Sorghum or 2 cowpeas, 2 Euonymus).
- 11. Check cages daily for mortality, health of plants, plant pest infestation, and maturity of insects.
- 12. Collect eggs from adult cages.
- 13. Feeding plants = Cowpea, chrysanthemum, sunflower Oviposition plants = Sorghum, euonymus

Egg Collection Techniques:

- 1. Examine each leaf on plant for egg masses. If any are found on cowpea or sorghum gently tap on stem of plant or leaf to dislodge sharpshooters. After sharpshooters are off of the plant remove it from the cage. On euonymus carefully pluck the leaf from the plant leaving as much stem as possible, Place leaf into plastic bag with damp paper towel.
- 2. Using a black Sharpie marker write the number under each egg mass and put the total count of the egg masses and the number of eggs in the egg mass on the label of the pot.

- 3. Place the plant on the holding table for three days if the eggs are going to be processed and put back into the colony.
- 4. If eggs are going to be used for wasp production set up in sting cages or store in cold room until they can be used.
- 5. Carefully remove leaves from plants and process. (See Lab Rearing Techniques)
- 6. Check soil to make sure plant has enough moisture.
- 7. Trade plant if old or unhealthy.
- 8. Trim plant of flowers, bean pods, runners, and dead or dying leaves. (Brown or yellow) Pinch tops of cowpea plants if they are growing too large.
- 9. Check number of insects in cage and combine with another cage if less than 50 bugs, unless cage is still producing well.
- 10. Clean bottom of cages, removing dead insects and debris.
- 11. Check for pests.
- 12. Fill out Egg Collection Data Sheets.